to be an ideal matrix for the seeding with vascular cells, its poor biomechanical properties have imitated their use. In our present study we evaluate a novel technique for the engineering of fibrin-based bioartificial vascular segments and report on first in vivo results.

Methods: The manufacturing process is based on the use of a custom-made rotating casting mould and the Vivostat-system for the separation and application of a fibrin precipitation from blood or plasma. Using this technique from 100 mL of blood 10 cm long tubular fibrin segments with an inner diameter of 5 mm were generated. With the optimized process generation of tubular fibrin segments was done immediately prior to implantation. To achieve antithrombogenicity the segments were seeded during the manufacturing process with endothelial and smooth muscle cells, which were isolated from the recipient's blood 4 weeks before and expanded in vitro. 6—8 cm long segments of the carotid artery of sheep were replaced by bioartificial vascular segments ($n = 6$), which were explanted after 1 and 6 months, respectively.

Results: The centrifugal force resulting from the rotation of the mould enhanced the cross-linking of thereby compacted fibrin fibrils and resulted in an up to 10-fold increase of the stability of the fibrin matrix. Using the optimized setting, autologous bioartificial vascular segments were generated within 1 hour prior to implantation. Whereas one segment ruptured immediately after implantation, after 1 month 3 of the remaining 5 segments were patent, 2 were closed due to dissection. 1 of the 3 patent segments was explanted at 1 month and the other 2 at 6 months after implantation. Subjected to the body's remodelling mechanisms in vivo, the segments showed an increasing at least high structural similarity to a native artery after explantation at 6 months.

Conclusion: Although a further optimization regarding biomechanical stability and antithrombogenicity is needed, the results of this study confirm that with the developed technique bioartificial small calibre vascular segments can be generated on demand immediately prior to implantation.

Electrospun Produced Small Diameter Vascular Grafts: Modification of Physical Properties and Assessment of Biocompatibility

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Introduction: Arteries more than 6 mm are efficiently replaced by allo- auto- xeno- or synthetic grafts whereas available grafts of small diameter arteries are obstructed by aneurisms or stenosis. The scaffolds produced by electrospinning represent convenient material for small diameter vascular grafts engineering but increasing flexural strength, kinked resistance, resistance to dishevel and suture retention as well as modification of the porosity, filling grafts with adequate cells are the limitations which should be overcome.

Methods: 3D matrixes (18 × 3 mm) or vascular grafts (i.d. 2 mm) were electrospun produced (EP) using NF-103 setup from polycaprolactone (PCL), nylon 6, polylactic-co-glicolic acid 50:50 (PLGA) and their mixtures with BSA or gelatin in 1,1,1,3,3,3-hexafluoroisopropanol. 2 MeV ILU-6 electron accelerator was used for 3D matrixes electron beam irradiation in doses 25 ÷ 150 kGy. Mechanical strength/structure were tested using Zwick/Roell Z100 testing machine/JSM-6460 LV scanning electron microscopy (SEM), porosity was tested as described in ISO 7198-98. Human primary endothelial cells (HUVEC) and gingival fibroblasts (HGF) were used to check in vitro biocompatibility by means of Axiovert 200 fluorescent microscopy (FM). Vascular grafts were implanted in Wistar rat's abdominal aorta; intravital MRI using BioSpec 117/16USR, histochemical or survey light/fluorescent microscopy (Discovery V12) were used to evaluate grafts functioning.

Results: 3D matrixes/vascular grafts from synthetic polymers or protein/polymer mix were EP. Supplementation of 5% gelatin into PCL increase proportional limit (PL) up to 60%, Young modulus to 50% and yield stress up to 45%. Irradiation increase PL of PCL twice in depend from the irradiation dosage, decrease stability of PLGA-matrix and the efficacy of protein release from mixed matrixes.

Especially produced 5—10 micron inner layer decrease permeability of the vascular grafts from ∼19 ± 3 ml to 0,5 ml. The data of SEM/FM demonstrate that irradiation does not interfere with adherence, viability and efficacy of proliferation of both HGF and HUVEC on 3D matrixes. Intravital functioning of vascular grafts using MRI, histochemical and survey light/fluorescent microscopy demonstrate normal functioning of the grafts in vivo.

Conclusion: Irradiation of electrospun produced matrixes was show to be a useful instrument to increase mechanic/chemical properties of the vascular grafts including introduction of stiffening elements (electron beam do not penetrate through less than one mm of steel) and/or their sterilization. In vitro and in vivo study demonstrates that the vascular grafts represent an efficient vascular prosthesis for reconstitution of small diameter blood vessels.

Wall Shear Stress Distribution in the Thoracic Aorta Using 4D MR Imaging: Potential Implications for Aneurysm Formation in Type B Dissection

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Introduction: Mechanical shear forces induced by blood flow play an important role in the process of vascular remodeling. Altered flow characteristics with regionally varying wall shear stress (WSS) have been demonstrated to correlate with the development of aneurysm formation using magnetic resonance (MR) imaging. The aim of this

study was to assess the distribution and regional differences in WSS in thoracic aortic dissection to further understand the process of aneurysm formation in this condition.
Methods: Ten patients with type B aortic dissection were examined by flow-sensitive four-dimensional (4D) MR imaging. Measurements of WSS (magnitude as well as axial and circumferential components) were taken in the four aortic segments: ascending aorta, arch, and proximal and distal descending thoracic aorta.
Results: The median time-averaged WSS in the true lumen was 0.09 N/m^2 (range 0.047–0.31 N/m^2), which was significantly greater than the false lumen 0.05 N/m^2 (0.02–0.17 N/m^2, $p =$ 0.0005). The highest overall WSS in the true lumen was found in the distal descending aorta (0.11 N/m^2 (0.055–0.31 N/m^2) and this was significantly greater than other aortic segments ($p = 0.02$). In the false lumen, peak values were seen in the proximal descending aorta just after the origin of the left subclavian artery (0.06 N/m^2 (0.024–0.17 N/m^2), and circumferential WSS (0.044 N/m^2 (0.016–0.14 N/m^2) was thirteen times greater than axial component of WSS.
Conclusion: Flow-sensitive 4D MR imaging is able to quantify the distribution of WSS in the entire thoracic aorta in patients with aortic dissection. Marked regional differences in WSS exist, which may help explain why specific segments of the aorta dilate while others remain stable over time in this condition.

A Randomised Controlled Trial of Supervised Exercise Regimens and their Impact on Walking Performance, Skeletal Muscle Mass and Calpain Activity in Patients with Intermittent Claudication

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Introduction: Supervised exercise training (SET) is recommended for patients with intermittent claudication (IC). The optimal exercise program hasn't been identified and the potential adverse effects of exercise on these patients warrant consideration. Calpain proteases have been linked with tissue atrophy following ischaemia-reperfusion injury. High calpain activity may therefore cause muscle wasting in claudicants undergoing SET and skeletal muscle mass (SMM) is integral to healthy ageing. This study assesses the impact of A: treadmill based SET alone; or B: combined with resistance training on pain-free walking distance (PFWD), SMM and calpain activity.
Methods: 35 patients with IC randomised to 12 weeks of treadmill only SET (Group A) or combined treadmill and lower-limb resistance SET (Group B). PFWD via 6-minute walk test, SMM via dual energy x-ray absorptiometry and calpain activity via biopsies of gastrocnemius muscles were analysed.
Results: Intention to treat analyses revealed PFWD improved within Group A (160 m to 204 m, $p = 0.03$) but not Group B (181 m to 188 m, $p = 0.82$), no between group difference, $p = 0.42$. Calpain activity increased within Group A (1.62 × 105 FU to 2.21 × 105 FU, $p = 0.05$) but not Group B, no between group difference, $p = 0.09$. SMM decreased within Group A (−250 g, $p = 0.11$) and increased in Group B (210 g, $p = 0.38$), $p = 0.10$ between groups. Similar trends were evident for per protocol analyses but additionally change in SMM was significantly different between groups ($p = 0.04$).
Conclusion: Neither exercise regimen was superior in terms of walking performance. Further work is required to investigate the impact of the calpain system on SMM in claudicants undertaking SET.

Inhibition of Individual 14q32 MicroRNAs Drastically Increases Neovascularization and Blood Flow Recovery after Ischemia

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Introduction: Neovascularization, i.e. angiogenesis and arteriogenesis, is a multifactorial process. As microRNAs can regulate expression of up to several hundred target genes, we hypothesized that specific microRNAs may target not just single aspects of neovascularization, but neovascularization as a whole. We set out to identify microRNAs that target genes in all pathways of neovascularization. Using www.targetscan.org, we performed a reverse target prediction on a set of 197 genes involved in neovascularization. We found enrichment of binding sites for 27 microRNAs in a single microRNA gene cluster on the long arm of human chromosome 14. MicroArray analyses showed that 14q32 microRNAs were down-regulated during effective neovascularization in mice subjected to single femoral artery ligation.
Methods: Gene Silencing Oligonucleotides (GSOs), were injected (1 mg/mouse) to inhibit four 14q32 microRNAs, miR-487b, miR-494, miR-329 and miR-495, one day prior to double ligation of the femoral artery. Blood flow recovery was followed by Laser Doppler Perfusion Imaging.
Results: All 4 GSOs clearly improved blood flow recovery after ischemia. Mice treated with GSO-495 or GSO-329 showed increased perfusion already after 3 days (30% perfusion vs. 15% in control animals) and those treated with GSO-329 showed a remarkable full recovery of perfusion after 7 days (vs. 60% perfusion in control animals). In vivo arteriogenesis was enhanced as 3-fold increased collateral artery diameters were observed in adductor muscles of GSO-treated mice. Simultaneously, in vivo angiogenesis was also enhanced as we observed up to 10-fold increased capillary densities in the ischemic soleus muscles of GSO-treated mice. Furthermore, in vitro treatment with GSO-329, GSO-495 and GSO-487b led to increased proliferation of primary human arterial endothelial cells whereas treatment with GSO-494 led to increased proliferation of primary human arterial fibroblasts.
Conclusion: Inhibition of 14q32 microRNAs leads to drastic increases in post-ischemic blood flow recovery in vivo via stimulation of both arteriogenesis and angiogenesis. In